Answer to letter of Aug 28

September 15, 1945

Dr. Van R. Potter
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Madison, Wisconsin

Dear Van;

Please pardon the long delay in answering your letter. It caught me at a very awkward time. Our new youngster (a boy) had arrived a few days previously and I was kept busy trying to keep our older boy happy. Besides our course for the med students was ending which is always a hectic time. In the midst of all this I had to keep the work going so that my technician wouldn't die of boredom (she didn't). I am just about now, on my way to regaining my sanity.

Now to try to answer the questions you raised in your letter. It seems to me that you are perfectly safe in assuming that the assayed activity is a valid measure of the enzymatic content. That these activities would be proportional to the relative amounts of substrate actually metabolized would be true however only under the following assumptions; (1) The enzyme assayed is the only metabolic pathway for succinate. (2) You are dealing with an enzymatically balanced (non-transforming) cell.

To give a trivial example which points up the necessity of making the second assumption we may imagine galactozymase assays on a galactose adapted cell no longer in contact with galactose. Under certain conditions I can maintain the enzyme almost indefinitely in the absence of its substrate. Thus conditions can exist where non-functioning enzymes can persist in measurable amounts and where activity measurements bear no relation to the amount of substrate metabolized via the assayed enzyme. It is not unlikely however that since you are dealing with tissue analysis (i.e. cells which have been around a long time and have had ample opportunity to get rid of non-functional enzymes) you need not concern yourself too much about the necessity of making assumption (2) altho I think it would be of value to explicitly state it. That would leave you a way out.

As to the data you request; some time ago I tried to obtain such data on succinoxidase with one of my yeast strains. I found that this particular enzyme would not form in the absence of an external source of nitrogen, i.e., it could not, unlike other enzymes in this strain, compete successfully with the existent enzymes for the cellular source of protiens. With an external source of nitrogen the level of enzyme activity becomes independent of substrate level beyond 0.2 %. This is the usual picture obtained and is not surprising in view of the general nature of substrate concentration-rate curves. The level of activity obtained is a $l_{\frac{1}{2}}$ of 2.1 before adaptation and 48 after the synthesis is complete. You will be interested to note that the $b_{\frac{1}{2}}$ with respect to glucose is 90.3 and is not detectably different in the succinate-adapted and non-adapted cells.

I should like to emphasize that this picture is true only for one strain and I don't know how general it is as yet. We are going to get further data on this point since succinoxidase is one of the enzymes selected for our study of competitive interaction during enzyme formation. It is of particular interest for our program since it represents one of

the extreme cases of an enzyme which must draw on external nitrogen sources to establish itself in the cytoplasm. It is also an extremely unstable enzyme even in the presence of its substrate if external nitrogen is withdrawn, which fits very well into the picture, of stability depending on competitive interaction in a metabolizing cell.

Should you wish to play around with it yourself, don't hesitate to do so and if you want some of our strains to do the job I would be happy to send them along.

Your comment re the Zoo department, 'fear of boat rocking' confirmed my own conclusions as to their probable reaction. Altho I know such fears are groundless there is unfortunately little I can do to dispel them.

By the way, I found out quite a bit about Huskins. Although I was unaware of it at the time I last saw you, I happen to know two very good friends of his, Dr. Edgar Anderson and Dr. L.J. Stadler. Both of them assured me he was a fine fellow who had a lot of ability and would not likely be one to try to organize anybody out of independent existence. He did a fine job at McGill in building up a Genetics department from scratch. I would by all means go to see him as he probably got much to contribute. On the basis of the discussion I had with Dr. Anderson I shall sendhim my reprints and try to get acquainted myself.

I am sending you reprints of that competition paper which you saw in galley proof. Peculiarly enough, the Bethesda cancer group got quite excited about this paper and wrote me some very nice fan mail.

This letter is getting too long and so I will save the rest for a later effort. I have some beautiful data now on competitive interaction between enzymes and most important involving some components of the glucozymase system which makes the whole phenomenon more general and makes me very happy.

Sincerely yours,

S. Spiegelman

P.S. Stowell is scared stiff that at the forthcoming meeting you will pulverize him with your biochemical erudition. I tried to calm his fears (which apparently arose via a correspondence between you two) and assured him you were a very gentle soul who didn't eat babies for breakfast only energy rich phosphate bonds.